

# TransStart® FastPfu Fly DNA Polymerase

Please read the manual carefully before use.

Cat. No. AP231

Version No. Version 3.0 Concentration: 2.5 units/µl Storage: at -20°C for two years

Description

TransStart® FastPfu Fly DNA Polymerase is a hot start, high-fidelity and high processivity DNA Polymerase used for fast PCR. Compared with TransStart® FastPfu DNA Polymerase, TransStart® FastPfu Fly DNA Polymerase has higher extension rate (6kb/min vs 4kb/min), featuring higher amplification efficiency, higher yield, higher fidelity and higher specificity.2×TransStart® FastPfu Fly Reaction Mix already contains dNTPs. When DNA is amplified, just add template, primer, water and TransStart® FastPfu Fly DNA Polymerase. The Reaction can be carried out with the concentration of Reaction Mix being 1×. PCR products are blunt end and can be cloned directly into pEASY®-Blunt series of vectors.

- Offers 108-fold fidelity as compared to *EasyTaq*® DNA Polymerase.
- PCR products can be directly cloned into  $pEASY^{\mathbb{R}}$ -Blunt vectors.
- Amplification of genomic DNA fragment up to 15 kb.
- Amplification of plasmid DNA fragment up to 20 kb.

#### Features

- Hot start, high specificity.
- High amplification efficiency.
- Fast and high fidelity.
- High sensitivity.
- High production.

#### **Applications**

- Complex templates, GC/AT-rich templates PCR
- Ultra high fidelity and fast PCR, blunt end cloning, site-directed mutagenesis.
- Long fragment amplification.

#### Kit Contents

Component	AP231-21-V3	AP231-22-V3	AP231-23-V3
TransStart® FastPfu Fly DNA Polymerase	250 U×1	500 U×1	500 U×6
2×TransStart® FastPfu Fly Reacti	1ml×3	1 ml×6	1ml×36
50 mM MgSO <sub>4</sub>	200 μl×1	400 μl×1	1ml×1
6×DNA Loading Buffer	500 μl×1	1ml×1	1ml×2
PCR Stimulant	200 μl×1	400 μl×1	1ml×1





## Reaction Components(50 µl reaction volumes)

Component	Volume	<b>Final Concentration</b>
Template	Variable	As required
Forward Primer (10 µM)	1 μl	0.2 μΜ
Reverse Primer (10 μM)	1 μ1	0.2 μΜ
2×TransStart® FastPfu Fly Reaction Mix	25 µl	1×
TransStart® FastPfu Fly DNA Polymerase	1 μl	2.5 units
Nuclease-free Water	Variable	-
Total volume	50 µl	-

# Optimized parameters (50 µl reaction volumes)

Template	Input
Genomic DNA	10-500 ng
Plasmid DNA	1-30 ng
cDNA	1-2 μl cDNA from RT reaction (50-500 ng RNA for RT reaction)

### **PCR**

Number of Cycles	Temperature	Time
1 cycle	95°C	2 min
30-35 cycles	95°C	20 sec
	Tm-5°C	20 sec
	72°C	6 kb/min
1 cycle	72°C	5 min

# **PCR Stimulant**

PCR Stimulant is used to optimize the amplification of complex templates or high GC/AT templates. The amplification of the Pfu series of enzymes is enhanced significantly. The concentration of the storage solution is  $5\times$ , and the concentration of the working solution can be adjusted between  $0.5\times-2.0\times$ 

## Notes

- For GC-rich templates, the recommended denaturation temperature is 98°C
- It is recommended to add TransStart® FastPfu Fly DNA Polymerase to the reaction system in the last step.
- If 2×TransStart® FastPfu Fly Reaction Mix has a small amount of precipitation after thawing, please heat it in a 37°C water bath and mix it for use.